

U.S. Army Center for Health Promotion
and Preventive Medicine

**Wildlife Toxicity Assessment for
NITROGLYCERIN (NG)**

NOVEMBER 2001

Prepared by
Health Effects Research Program
Environmental Health Risk Assessment Program

USACHPPM Document No: 37-EJ-1138-01F
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Department of the Army
U.S. Army Center for Health Promotion and Preventive Medicine

Wildlife Toxicity Assessment for Nitroglycerin

CAS No. 2691-41-0

October 2001

1. INTRODUCTION

This Wildlife Toxicity Assessment (WTA) is based on a thorough review of the scientific literature regarding the toxicological characteristics of nitroglycerin that may pertain to the health of wildlife (mammals, birds, reptiles and amphibians) exposed to the substance. The protocol for the performance of this assessment is documented in the U.S. Army Center for Health Promotion and Preventive Medicine Technical Guide 254, the *Standard Practice for Wildlife Toxicity Reference Values* (USACHPPM 2000). This document is designed to support ecological risk assessment activities.

2. TOXICITY PROFILE

2.1 Literature Review

Nitroglycerin (also known as trinitroglycerol or TNG) is produced for use as a component of propellants and explosives, and as a pharmaceutical agent. Nitroglycerin is a vasodilator widely used by the medical community in the treatment of angina and other cardiovascular pathology (Gilman et al. 1990, Burrows et al. 1989). There is an extensive body of literature related to the medical uses of nitroglycerin. However, very few of these studies were found relevant to a WTA and the development of wildlife toxicity values.

Given the common military use of NG, several pertinent studies were found in U.S. Army and related sources. These military-related studies, and subsequent reports, were found through TOXLINE, Toxicology, Occupational Medicine Environmental Series (TOMES) and Defense Technical Information Center (DTIC) searches. In addition, relevant studies were found through traditional cross-referencing techniques and through individual queries to project investigators within the Army. Several databases were searched; the details of these searches are found in Appendix A.

Table 1. Table of Physical / Chemical Properties of Nitroglycerin

CAS:	55-63-0
Molecular weight	227.09
Color	Pale yellow
State	Viscous liquid; triclinic or rhombic crystals below melting point
Melting point	13.5°C/2.8°C (labile form)
Boiling point	Apparently boils by decomposing rapidly at temperatures above 145°C; Explodes at 218-256°C
Taste	Sweet, burning taste
Solubility	1800 mg/L @ 25°C; soluble in acetone, benzene, and chloroform.
Partition coefficients	
Log K _{OW}	1.62
Log K _{OC}	1.51 estimated
Vapor pressure (at 20°C)	2.5E-04 mm Hg
Henry's Law constant (at 20°C)	Between 3.3 and 0.06 torr-l/mole
Odor	No information
Conversion factors	1 ppm = 9.29 mg/m ³ 1 mg/m ³ = 0.108 ppm

Sources: USEPA 1992, HSDB 2000, Lyman, W.J. et al. 1985, ACGIH 1991

2.2 Environmental Fate and Transport

Nitroglycerin may be released to the environment from its production and use as a component of propellants and explosives and as a pharmaceutical compound. Wastewater discharges from the manufacture of commercial dynamite preparations, military explosives, and other production sources may contain NG. The relatively high solubility of NG in water (1800 mg/L @ 20°C) suggests that environmentally significant concentrations may be dissolved in waste rinse water and be expected to remain in the water column (USEPA 1992). Neat NG does not polymerize and exists as a stable solid in the form of dipyramidal crystals at temperatures below 13.5°C. Liquid NG begins to decompose at 50 to 60°C, and at 145°C decomposition is so rapid the liquid appears to boil. At 256°C NG explodes spontaneously (USEPA 1992).

Both abiotic and biotic processes influence the fate of NG released to the environment. Physical and chemical degradation of NG is generally slow (Smith 1986). The photolytic half-life of NG in pure water was estimated to be 5 days (Spanggord et al. 1980a), and the hydrolysis half-life of NG at normal environmental temperatures is predicted to be more than 1 year at pH 3 to 8 (Spanggord et al. 1980b). An alkaline environment significantly decreases the hydrolysis half-life of NG (Spanggord et al. 1980b).

The kinetics and products formed on hydrolysis of NG and each of the isomeric dinitroglycerols (DNGs) and mononitroglycerols (MNGs) have been extensively studied. Chemical hydrolysis does not follow the stepwise successively slower pathway found for microbial systems. Rather, NG hydrolyzes more slowly than the DNGs or MNGs to a complex mixture of products, notably nitrate, nitrite, and oxalate. 1,3-DNG is hydrolyzed to glycidyl nitrate and 1,2-DNG isomerizes to 1,3-DNG to form a similar product on hydrolysis. 2-MNG isomerized to 1-MNG before hydrolysis; the only product identified was nitrate (Capellos et al. 1984).

Nitrate and nitrite are highly mobile in soil and water and microbes can convert nitrate to nitrite, which can potentially cause methemoglobinemia in mammals. Plants may accumulate nitrates and ingestion of plant material by ruminants (rumen microorganisms reduce nitrate to nitrite) may also result in methemoglobinemia (Cockerham 1994). However, the production of hydrolysis products from NG is not expected to be of concern to wildlife because the reaction will not proceed at any appreciable rate outside of an environmentally extreme alkaline environment.

Contrary to some earlier reports that it was recalcitrant to biodegradation, NG proved to be readily biodegradable in batch and continuous tests. Breakdown of NG was found to take place in stages via the isomeric di- and mononitrates, with each successive step proceeding at a slower rate (Wendt et al. 1978, Walker and Kaplan 1992). It was found that NG is not suitable as a source of carbon and nitrogen so nutrients are essential. It was speculated that earlier experiments where NG did not biodegrade were conducted using NG concentrations that were toxic to the microorganisms (Wendt et al. 1978, Smith 1986, Burrows et al. 1989). In the environment, NG would likely be biotransformed through a series of successive denitration steps, and the products mineralized by biological systems and incorporated into the biomass (Walker and Kaplan 1992).

If released on land, NG may readily leach into the soil. Few experimental data are available on its fate in the soil but biodegradation and hydrolysis under alkaline conditions are thought to occur. Few data are available on the fate of NG released in aquatic environments; however, environmental degradation of NG appears to occur primarily through biodegradation and photolysis. If released to the atmosphere, NG will probably be in the form of an aerosol and be subject to gravitational settling and scouring by rain. Photolysis is considered a possibility but data are lacking. The Henry's Law constant calculated from

reported water solubilities and vapor pressures range between 3.3 and 0.06 torr-l/mole, resulting in a half-life for volatilization from water of about 3000 days. Therefore, physical transport from aqueous systems should be relatively unimportant. Nitroglycerin is relatively soluble in water (1800 mg/L at 25°C) and therefore adsorption to soil and sediment and bioconcentration in aquatic organisms should not be appreciable (HSDB 2000, Smith 1986, Spangford et al. 1980b). Chemical/physical properties of nitroglycerin are presented in Table 1.

2.3 Summary Of Mammalian Toxicology

Nitroglycerin has profound effects on systemic as well as cardiac microcirculation. Its actions are mediated by stimulation of soluble guanylate cyclase in vascular smooth muscle cells. Long-term industrial exposure to NG has been associated with withdrawal symptoms and sudden death from cardiovascular accidents (Klaassen 1996).

Nitroglycerin is rapidly absorbed, rapidly and widely distributed, and rapidly metabolized and eliminated in both laboratory animals and humans. Metabolism appears to occur in both hepatic and extrahepatic tissues via stepwise denitification; elimination is primarily in the urine and expired air. Absorption is somewhat less in mice than other species (Smith et al. 1986).

Urinary metabolites in most species consisted largely of free MNGs, glycerol, and other polar metabolites including glucuronides, while TNG and free DNGs were excreted only in small amounts. Mice excreted only small amounts of free MNG and DNG- and MNG-glucuronides indicating the relatively complete biotransformation in this animal species (USEPA 1992).

Nitroglycerin is absorbed through intact skin in amounts sufficient to cause vasodilation. In humans the most prominent manifestations of NG toxicity are severe headaches and adverse cardiovascular effects, including organic nitrate dependence in the case of chronic exposure (Gilman et al. 1990). In animals, the adverse effect most often observed after administration of NG at high dosage levels is decreased weight gain (related to decreased food consumption); effects were also seen in the liver (lesions), blood (methemoglobinemia), and testes (lesions and aspermatogenesis) (USEPA 1992).

Nitroglycerin has not been shown to be genotoxic in either *in vivo* or *in vitro* studies. Developmental and reproductive studies in animals have failed to demonstrate that NG is a teratogen. However, exposure to high concentrations of NG can result in testicular lesions and male infertility, and delayed development of offspring (Smith 1986).

2.3.1 Mammalian Oral Toxicity

2.3.1.1 Mammalian Oral Toxicity – Acute

The acute toxicity of NG in mammals is moderate. The oral LD₅₀ was reported to be approximately 500-900 mg/kg in rats and 500-1200 mg/kg in mice (Lee et al. 1975, Oketani et al. 1982). Differences in the acute toxicity of NG between sexes or among species appeared insignificant or minor.

Charles River CD rats and Albino Swiss mice were administered NG in lactose/peanut oil by oral gavage and survivors observed for 14 days. The oral LD₅₀ was determined to be 822 and 884 mg/kg in male and female rats, respectively, and 1188 and 1055 mg/kg in male and female mice, respectively. All the animals became cyanotic and ataxic within 1 hour of dosing, and had very pale extremities and depressed respiration. Animals usually died within 5-6 hours of dosing; no gross pathology was observed in the animals that died. Survivors generally recovered within 24 hours (Lee et al. 1975). The number of male and female animals used in these studies was not provided.

Oketani et al. (1982) administered NG in propylene glycol to S1c:dd mice and S1c:SD rats by oral gavage and observed the surviving animals for 7 days. Ten animals per sex were used in these studies. The LD₅₀ was 525 and 540 mg/kg for male and female rats, respectively, and 550 and 500 mg/kg for male and female mice, respectively. Deaths occurred within 48 hours and survivors recovered within 48-72 hours. There were no notable findings at autopsy (Oketani et al. 1982).

Four groups of adult beagle dogs, each consisting of two males and two females were given 25, 50, 100 or 200 mg/kg/day NG in capsules for 5 days. A transient and dose-related severe methemoglobinemia was observed. The 100 and 200 mg/kg/day group animals had cyanosis lasting for several hours. High dose dogs also exhibited decreased activity. Because of the severity of effects and to study any protective effect on methemoglobin formation, the high dose animals were given 3 mg/kg methylene blue intravenously (to induce methemoglobin formation) on the 3rd day and treatment was discontinued thereafter. The two low dose groups had transient methemoglobinemia and no adverse clinical effects (Lee et al. 1977, Ellis et al. 1984).

2.3.1.2 Mammalian Oral Toxicity – Subchronic

Thirteen-week oral studies were conducted in dogs, rats, and mice as described in the following paragraphs (Lee et al. 1977, Ellis et al. 1984). Because no adverse effects were observed during the first several weeks of the 13-week studies, doses were increased 5-fold for dogs, rats, and mice starting the 5th, 6th, and 4th week, respectively.

Healthy young adult beagle dogs, 4 males and 4 females per treatment group, were given 0, 0.01, 0.1 or 1 mg/kg/day NG in capsules for 4 weeks, then 0, 0.05, 0.5 and 5 mg/kg/day for 9 more weeks. No

adverse effects were seen at any dose (Lee et al. 1977, Ellis et al. 1984). The no observed adverse effect level (NOAEL) was 5.0 mg/kg/day.

CD-1 mice, 16 male and 16 female per treatment group, were fed 0.001, 0.01 or 0.1% NG in diet (intakes calculated to be 1.3/1.3 m/f, 11.5/10.9 m/f, 107/95 mf mg/kg/day) for 3 weeks, then 0.005, 0.05 and 0.5% (intakes calculated to be 6.4/6.9, 60.2/58.7m/f, 608/561m/f mg/kg/day) for 10 more weeks. Treated mice had mild extramedullary hematopoiesis in the liver and spleen that did not appear dose-related. No other adverse effects were observed at any dose level (Lee et al. 1977, Ellis et al. 1984). The NOAEL was 608 and 561 mg/kg/day in males and females, respectively.

CD rats, 16 males and 16 females per treatment group, were fed 0.001, 0.01, or 0.1% NG in diet (intakes were calculated to be 0.8 and 0.9, 6.0 and 6.4, 59 and 59 mg/kg/day in males and females, respectively) for 5 weeks then 0.005, 0.05 and 0.5% (intakes were calculated to be 2.6 and 3.1, 24.5 and 26.5, 230 and 234 mg/kg/day in males and females, respectively) for 8 more weeks. Reversible decreases in food consumption, weight gain, and increased SGOT (aspartate aminotransferase) levels (enzymes to detect liver damage) were observed after the increase in dosage in the high-dose group; no other toxicologically significant findings were observed. No adverse effects were observed in any other dose groups. The lowest observed adverse effect level (LOAEL) was 230 and 234 mg/kg/day for decreased weight gain and food consumption in males and females, respectively. The NOAEL was 24.5 (males) and 26.5 (females) mg/kg/day.

To assess the effects of higher doses of NG, CD rats, 3 males and 3 females (4 males and 4 female controls) were fed 2.5% NG in diet (overall average intake of 1406 or 1416 mg/kg/day, m/f) for 13 weeks (Lee et al. 1977, Ellis et al. 1984). Dosing for males and females was initiated at 1176 and 1076 mg/kg/day and increased to 1588 or 1773 mg/kg/day, respectively, during the last 5 weeks. A lactose control group was also included with 5 males and 5 females. Animals receiving the NG experienced weight loss (weeks 4-8) and compensated anemia, but resumed gaining weight as feeding continued. Treated rats had altered blood chemistries and relative organ weights, hemosiderosis in the liver and spleen, and moderate to severe testicular degeneration and/or atrophy with severe and complete aspermatogenesis.

2.3.1.3 Mammalian Oral Toxicity – Chronic

Chronic exposure of laboratory animals to high NG concentrations resulted in adverse hematological and liver changes, and decreased body weight gain. Hepatocellular carcinomas as neoplastic nodules, and interstitial cell tumors of the testes were frequently observed in the high-dose group rats after 2-year exposures to NG (Ellis et al. 1978a, Ellis et al. 1984).

Four groups of 38 male and 38 female Charles River CD albino rats were fed diets containing 0, 0.01, 0.1 or 1% (average intakes of 0, 3.04 and 3.99, 31.5 and 38.1; 363 and 436 mg/kg/day in males and females, respectively) NG in their diet for 2 years (Ellis et al. 1978a, Ellis et al. 1984). No adverse effects were observed in any of the low dose rats. Mid-dose rats exhibited decreased weight gain in later months; males and females were about 60 and 30 g lighter than controls, respectively. Some rats fed 0.1% NG (31.0 (males) or 38.1 (females) mg/kg/day) had mild hepatic lesions (areas or foci of hepatocellular alteration that can develop into hepatocellular carcinomas). High-dose rats had decreased food consumption and weight gain, behavioral effects (decreased activity and failure to groom) compensated anemia with reticulocytosis, elevated serum transaminases, and methemoglobinemia, with some excessive pigmentation in the spleens and renal epithelium. After 1 year, 8 high-dose rats had cholangiofibrosis and some had neoplastic foci in the liver. At 2 years, all 13 surviving high-dose rats and 6/16 middle-dose rats had enlarged and grossly abnormal livers with severe cholangiofibrosis and hepatocellular carcinomas, some of which had metastasized to the lung. Interstitial tumors of the testes were observed in one-half the high dose males, in some leading to aspermatogenesis. A decrease in the naturally occurring pituitary chromophobe adenoma and mammary tumors increased the life-span, especially in the females (Ellis et al. 1978a, Dacre et al. 1980, Ellis et al. 1984). The identified NOAEL for this study was 3.04 and 3.99 mg/kg/day and the LOAEL was 31.5 and 38.1 mg/kg/day for decreased weight gain and enlarged, abnormal livers with cholangiofibrosis and hepatocellular carcinomas in males and females, respectively. The LOAEL for male reproductive effects was 363 mg/kg/day. The NOAEL for male reproductive effects was 31.5 mg/kg/day.

Four groups of 58 male and 58 female CD-1 mice were fed diets containing 0, 0.01, 0.1 or 1% (average intake of 0, 11.1 and 9.7, 114.6 and 96.4; 1022 and 1058 mg/kg/day for males and females, respectively) NG for 2 years (Ellis et al. 1978a, 1984, Dacre et al. 1980). No adverse effects were seen in the low- and mid-dose groups during the 24-month study. Decreased feed consumption and weight gain, and behavioral effects (decreased activity and failure to groom) were observed in the high-dose mice. After 1 year, high-dose animals had heme-derived pigment deposits in various organs and liver dysplasia. At 2 years, pigmentation in the liver with a lesser amount in the spleen and/or kidneys was observed in most high-dose and some middle-dose mice (Ellis et al. 1978a, 1984, Dacre et al. 1980). High-dose mice also had decreased weight gain, decreased grooming and methemoglobinemia and its sequelae (Ellis et al. 1978a, 1984). The NOAEL was 11.1 and 9.7 mg/kg/day; the LOAEL was 114.6 and 96.4 mg/kg/day in males and females, respectively, for pigment deposits in liver, spleen and/or kidneys.

Four groups of 6 male and 6 female beagle dogs were given capsules containing 0, 1, 5, or 25-mg/kg/day NG for 1 year. The only effect observed was an occasional dose-related incidence of transient

mild methemoglobinemia (less than 3%) in some dogs at all dose levels treated for 6 months or more (Ellis et al. 1978a, 1984). There were no accompanying effects on body weight, feed consumption, other hematological tests, clinical chemistry and histological examination. The NOAEL was identified as 25.0 mg/kg/day (Ellis et al. 1978a, 1984).

There were quantitative and qualitative differences in effects between species. Oral doses up to 20 mg/kg/day for 5 days or 25 mg/kg/day for 12 months produced only transient methemoglobinemia in dogs. In rats, lifetime feeding of 363 mg/kg/day (males) and 434 mg/kg/day (females) resulted in toxic effects on the liver and blood, and 31.5 or 38.1 mg/kg/day (males and females, respectively) resulted in mild effects on the liver of some animals. Mice were the least affected. Lifetime feeding of 115 mg/kg/day (males) and 96 mg/kg/day (females) resulted in a compensated anemia and some pigment deposits. The only effect common to all three species was transitory methemoglobinemia. Increased interstitial cell tumors of the testes occurred only in rats (Ellis et al. 1978a, 1984).

Suzuki et al. (1975) exposed C57BL/6Jms mice to NG in their drinking water for 18 months at estimated oral administered doses of 0, 1.5, or 6.2 mg/kg/day, and for 12 months at an estimated concentration of 58.1 mg/kg/day. Each treatment group consisted of approximately 50 animals of each sex. No treatment-related adverse effects, including body weight changes, were observed in any dose groups. The NOAEL for the 18-month study was 6.2 mg/kg/day. The NOAEL for the 12-month study was 58.1 mg/kg/day.

2.3.1.4 Mammalian Oral Toxicity – Other

Ellis et al. (1978a) also conducted a three-generation reproductive study in CD rats where the parental generation (F_0) received the same concentrations of NG as the ones in the chronic study for 6 months prior to mating (i.e., 0, 0.01, 0.1 or 1% NG). Matings consisted of 10 males and 20 females from each group for the F_0 generation. Twenty to 24 pups from the second litters were randomly chosen in equal numbers from each treatment group and maintained in each respective treatment. At 3 months old, each male was mated with a female from each group and again, only the second-generation offspring were selected for continued treatment. This was repeated until the animals from the 3rd generation (F_{3b} 's) were weaned. All offspring were evaluated for gross physical abnormalities, and the number of live and dead pups were recorded. Survival and body weights were recorded at 0, 4, and 21 days. Fertility in the F_1 and F_2 generation of high-dose males was severely impacted. These effects appeared to result from the decreased feed intake and consequent poor nutritional status of the females and decreased spermatogenesis (due to interstitial tumors) in the males. No other developmental or reproductive effects were identified (Ellis et al. 1978a).

Ellis et al. (1978a) conducted a series of cytogenic studies in dogs and rats in the chronic study. In addition, dominant lethal mutation studies were also conducted. There were no apparent NG-induced mutagenic effects in the cytogenetics analyses of cells from dogs and rats and in the dominant lethal mutation study in rats.

2.3.1.5 Studies Relevant for Mammalian TRV Development for Ingestion Exposures

The relevant studies for development of Toxicity Reference Values (TRVs) are the chronic and subchronic studies performed by Ellis et al. (1978a and 1984) and Lee et al. (1975 and 1977) in dogs, rats, and mice. These studies were conducted properly, containing the information necessary to evaluate the results, and reported results in a clear, logical, concise form. The methods used were consistent with other accepted protocols and contained the proper controls. These studies, therefore, meet or exceed the minimum quality requirements. The chronic studies were considered most relevant both because of the exposure duration and because the dose levels changed during the subchronic studies. This results in NOAEL and LOAEL values that are less certain. Acute studies provided additional information but were not considered applicable to the development of a TRV. The studies by Suzuki et al. (1975) and Oketani et al. (1982) were included as informational data points but were not used to derive a TRV, because these data were from a secondary reference and an English version of the original work could not be obtained. Table 2 summarizes the data from these studies and Figure 1 presents the data in a scatter diagram.

Table 2. Summary of Relevant Mammal Data

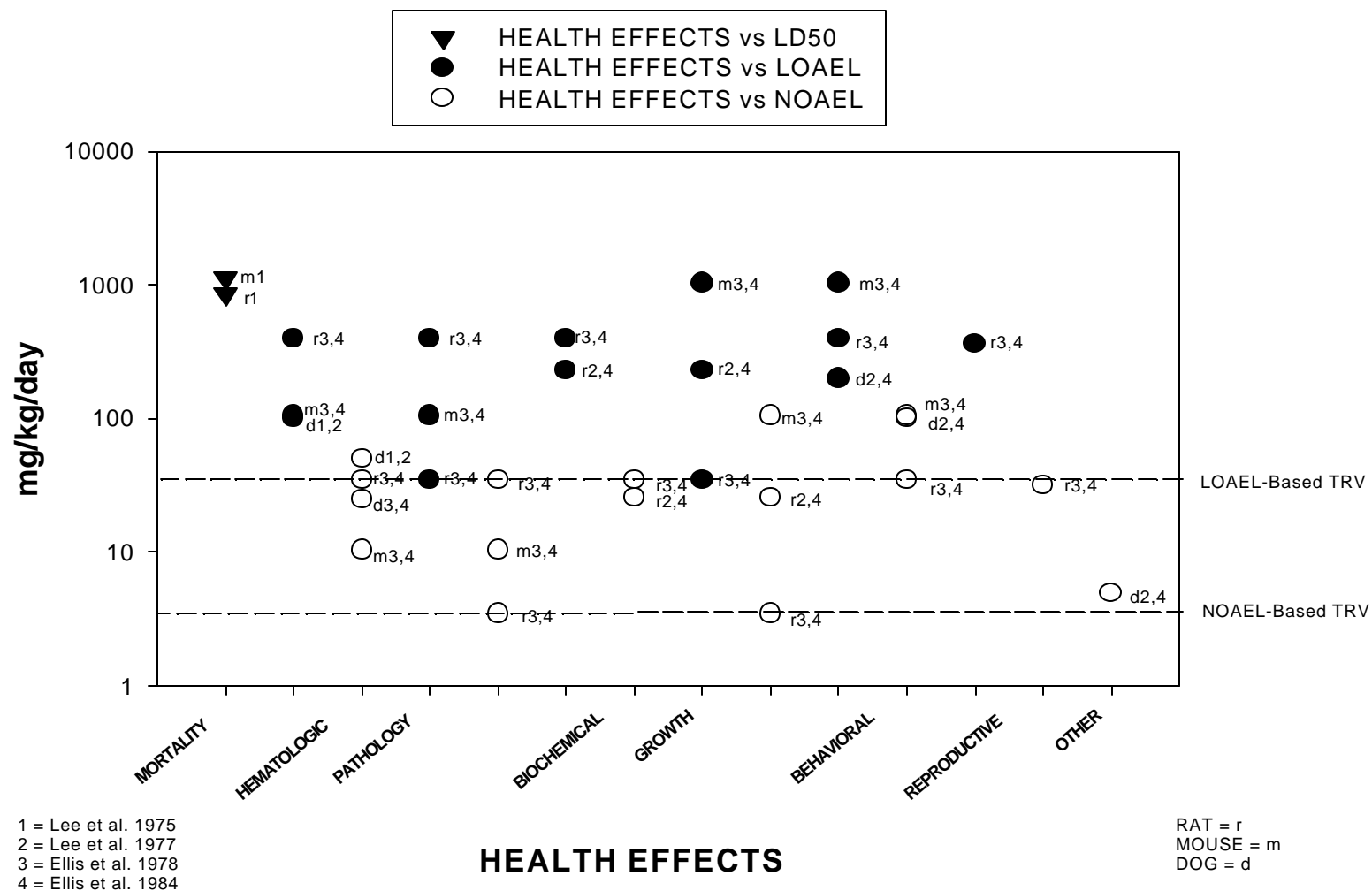
Study	Test Organism/Route	Test Duration	Test Results		
			NOAEL	LOAEL	Effects at LOAEL
Lee et al., 1975	Mouse/Albino Swiss/Oral in lactose/peanut oil	Single dose		LD50 1188 (M) 1055 (F)	Animals usually died within 5-6 hours; survivors recovered within 24 hours; no gross pathology. N = unknown
Lee et al., 1975	Rat/Charles River/Oral in lactose/peanut oil	Single dose		LD50 822 (M) 884(F)	Animals usually died within 5-6 hours; survivors recovered within 24 hours; no gross pathology. N = unknown
Lee et al. 1977 Ellis et al. 1984	Dogs/oral-capsule	5 days	50 mg/kg/day	100 mg/kg/day	Methemoglobinemia; Decreased activity. Cyanosis.
Lee et al. 1977 Ellis et al. 1984	Dog/oral/feed	13 weeks	5.0 g/kg/day	NA	0, 0.01, 0.1 or 1 mg/kg/day for 4 weeks, then 0, 0.05, 0.5 and 5 mg/kg/day for 9 more weeks. No adverse effects seen at any dose. Low number of animals.
Lee et al. 1977 Ellis et al. 1984	Mouse/oral/feed	13 weeks	608 (m) and 561 (f) mg/kg/day	NA	0.001, 0.01 or 0.1% in diet (intake of 1.3/1.3m/f, 11.5/10.9 m/f, 107/95 mf mg/kg/day) for 3 weeks, then 0.005, 0.05 and 0.5% (intake of 6.4/6.9, 60.2/58.7m/f, 608/561m/f mg/kg/day) for 10 more weeks. No treatment-related adverse effects at any dose. USEPA (1992) derived an LOAEL of 6.7 mg/kg/day based on mild hemosiderosis. Informational NOAEL, not considered for derivation of TRVs.

Study	Test Organism/Route	Test Duration	Test Results		
			NOAEL	LOAEL	Effects at LOAEL
Lee et al. 1977 Ellis et al. 1984	Rat/oral/feed	13 weeks	24.5 (m) and 26.5 (f) mg/kg/day	230 (m) and 234 (f) mg/kg/day	Decrease in weight gain and food consumption after increase in dosage in high dose group.
Lee et al. 1977 Ellis et al. 1984	Rat/oral/feed	13 weeks	NA	NA	Only one dose level. (2.5% ;1406 or 1416 mg/kg/day) - initial weight loss, altered blood chemistries, hemosiderosis in liver and spleen; testicular degeneration and aspermatogenesis.
Ellis et al. 1978a Dacre et al. 1980 Ellis et al. 1984	Dog/oral/capsule	12 months	5.0 mg/kg/day	NA	Mild methemoglobinemia (less than 3%, observed at all dose levels). USEPA 1992 did not derive NOAEL. Informational NOAEL, not considered for derivation of TRVs.
Ellis et al. 1978a Dacre et al. 1980 Ellis et al. 1984	Mouse/oral/ feed	2 years	11.10 (m) and 9.72 (f) mg/kg/day	114.6 (m) and 96.4 (f) mg/kg/day	Decreased weight gain, decreased grooming, hemosiderosis.
Ellis et al. 1978a Dacre et al. 1980 Ellis et al. 1984	Rat/Charles River/oral/ feed	2 years	3.04 (m) and 3.99 (f) mg/kg/day	31.5 (m) and 38.1(f) mg/kg/day	Reduced growth (weight loss); mild hepatic lesions like those seen in rats fed higher doses. No adverse effects were observed in the low dose rats.
Ellis et al. 1978a	Rat/Charles River/oral/ feed	3- Generation reproduction study	31.5 (m) and 38.1(f) mg/kg/day	363 (m) and 434 (f) mg/k/day	Infertility in high dose group (No effects in F ₀ , 1st mating of F ₁ produced 3 litters, only 1 F ₂ litter). Infertility due to males, testes ¼ size, no sperm in vaginal plugs, severe aspermatogenesis in F ₂ males.
Ellis et al. 1978a, 1984	Rat/Charles River/oral/ feed	2 years	31.5 mg/kg/day	363 mg/kg/day	Interstitial cell tumors of the testes with aspermatogenesis.

NA = not applicable

Figure 1

NITROGLYCERIN HEALTH EFFECTS IN MAMMALS



2.3.2 Mammalian Toxicity- Inhalation

No inhalation data for mammals were found.

2.3.3 Mammalian Toxicity- Dermal

Nitroglycerin was a very mild skin irritant but not an eye irritant in rabbits, and is a moderate sensitizer in guinea pigs (Lee et al. 1975). Nitroglycerin was readily absorbed through the skin of rhesus monkeys (Wester et al. 1983). No other relevant dermal toxicity data in mammals were found. However, the potential contribution of the dermal pathway to total exposure dose should be considered in the context of any risk assessment.

2.4 Summary of Avian Toxicology

Toxicological data for the effects of NG in avian species was not located. Ecotoxicological research on the effects of this compound in birds is recommended.

2.5 Summary of Amphibian Toxicology

Toxicological data for the effects of NG in amphibian species was not located. Ecotoxicological research on the effects of this compound in amphibians is recommended..

2.6 Summary of Reptilian Toxicology

Toxicological data for the effects of NG in reptilian species was not located. Ecotoxicological research on the effects of this compound in reptiles is recommended.

3. RECOMMENDED TOXICITY REFERENCE VALUES¹

3.1 Toxicity Reference Values for Mammals

3.1.1 TRVs for Ingestion Exposures for the Class Mammalia

Three species of mammals from two different orders were evaluated using dietary exposures of TNG. The data set contains studies that are comprehensive in the scope of potential effects and in study duration. Moreover, most studies were well documented and contain refined data from all observations. Thus, these studies are considered to be of acceptable quality to derive a TRV.

Most studies have consistent and corroborative findings. Incidences of indicators that suggest liver necrosis, red blood cell lysis, methemoglobinemia, adverse reproductive effects, and weight loss were reported within the same ranges. Many studies conducted at relatively low levels failed to find adverse effects. Although some findings (e.g., transient and reversible methemoglobinemia) have questionable biological and ecological significance, other effects (e.g., reduced male fertility) are clearly important. Given that most of these data follow a consistent pattern, and that the reproductive data are not appropriate for benchmark dose extrapolation, the NOAEL-LOAEL approach was used.

The NOAEL-based TRV was based on the chronic study by Ellis et al. (1978a) where no adverse effects in weight loss or hepatic lesions were observed. Therefore, the NOAEL was derived from the most sensitive species (rat) and sex tested (males) at 3 mg/kg/d.

The LOAEL-based TRV was derived from the original Ellis et al (1978a) work where at 31.5 mg/kg/d mild hepatic lesions and incidences of weight loss were found in chronically exposed male rats. Therefore, the LOAEL-based TRV of 32 mg/kg/d was selected. This value less than those where other endpoints (e.g., reproductive) were observed and therefore protective of other reported effects.

Because the relevant data, as presented in Section 2.3.1.5, satisfies the minimum data set requirement of the Standard Practice (USACHPPM 2000), no uncertainty factors are needed to select the TRVs.

The studies used to derive the LOAEL-based TRV were chronic and the results are consistent with those of other studies, thus this TRV is given a **High** degree of confidence. The NOAEL-based TRV was

¹TRVs are for screening purposes only and are not intended to be predictors of effects in field situations. Site specific conditions may justify adjustments of these values based on toxicity information relevant to specific assessment endpoints.

derived from a chronic study that evaluated consistent endpoints of toxicity. However, given the information present in other studies where higher doses were tested in between these of Ellis et al. where no adverse effects were observed, it is likely that the true NOAEL for many species of mammals is likely to be higher. Therefore, the NOAEL-based TRV is given a **Medium** degree of confidence. See Table 3 for a summary of mammalian TRVs.

Table 3. TRVs for the Class Mammalia

TRV	Dose	Confidence
NOAEL-based	3.0 mg/kg/d	Medium
LOAEL-based	32.0 mg/kg/d	High

3.1.2 TRVs for Ingestion Exposures for Mammalian Foraging Guilds

TRVs specific to particular guild associations (e.g., small herbivorous mammals) have not yet been derived.

3.1.3 TRVs for Inhalation Exposures for the Class Mammalia

At this time, no inhalation TRV can be derived for mammals due to insufficient data.

3.1.4 TRVs for Dermal Exposures for the Class Mammalia

At this time, no dermal TRV can be derived for mammals due to insufficient data. However, given that there is evidence that NG has irritant and sensitizing properties, and that dermal exposures have been identified as significant, the dermal route of exposure should be considered for risk assessment purposes.

3.2 Toxicity Reference Values for Birds

At this time no TRVs for birds can be derived due to the lack of data.

3.3 Toxicity Reference Values for Amphibians

At this time no TRVs for reptiles and amphibians can be derived due to the lack of data.

3.4 Toxicity Reference Values for Reptiles

At this time no TRVs for reptiles and reptiles can be derived due to the lack of data.

4. IMPORTANT RESEARCH NEEDS

The effects of NG have not been assessed in avian, amphibian, or reptilian animal systems. It is recommended that these effects be evaluated.

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APPENDIX

LITERATURE REVIEW

The following databases were searched using the following keywords:

TOXLINE & MEDLINE

Conditions: Two-word search; 1965 to present.

<i>Nitroglycerin and mammals</i> -	Nitroglycerin = 1938
	Mammals = 29485
	Combination = 448

Of these, 6 were appropriate and included.

<i>Nitroglycerin and birds</i> -	Nitroglycerin = 1938
	Birds = 12127
	Combination = 1

After review of the title, the single query result was not appropriate for this document.

<i>Nitroglycerin and wildlife</i> -	Nitroglycerin = 1938
	Wildlife = 12029
	Combination = 0

<i>Nitroglycerin and salamanders</i> -	Nitroglycerin = 1938
	Salamanders = 425
	Combination = 0

<i>Nitroglycerine and toads</i> -	Nitroglycerin = 911
	Toad = 411
	Combination = 0

Nitroglycerin and reptiles - Nitroglycerin = 911
Reptiles = 4886
Combination = 0

Nitroglycerin and snake - Nitroglycerin = 911
Snake = 5825
Combination = 0

BIOSIS

(Agricola and Biological Abstracts)

Conditions: One word search; 1984-1999.

Nitroglycerin = 381

Of these, none were appropriate for this document.

WORLD WILDLIFE

Conditions: One word search

Nitroglycerin = 0

STINET – DTIC

Conditions: one word search

Nitroglycerin = 25

Of these, 2 were considered appropriate and included

In addition, the USEPA Health Advisory for Nitroglycerin (1992) and the Water Quality Criteria for Nitroglycerin (ORNL, 1986) references were consulted and all relevant non-duplicate studies included. Several others, in Japanese, appeared relevant but were not translated due to time and budgetary constraints.

The TOMES database was also searched; most relevant toxicological articles were in Japanese or duplicates from searches above. General chemical and fate and transport information from the HSDB (1999) was included.